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THE ANTIBACTERIAL MOUTHWASH OF TAPAK LIMAN LEAVES EXTRACT (*Elephanthopus scaber L*) AGAINST *Streptococcus mutans*

Ifmaily [1]and Putri Rizki Fitriani[2]
[1]Sekolah Tinggi Ilmu Farmasi Indonesia (STIFI) Perintis Padang
[2]College student of Faculty of Medicine of Andalas University

Corresponding Author: Ifmaily, Sekolah Tinggi Ilmu Farmasi Indonesia (STIFI) Perintis Padang.
Email : ifmaily2c@gmail.com

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Abstract

Biodiversity Indonesia has been widely used as a treatment for Indonesian people. One of them is a plant of Tapak liman that efficacious antibacterial. The purpose of this research to know the formulation of the leaf extract of Tapak Liman which can be made as a antiseptic mouthwash and know the inhibiting power test of the leaf extract of the Tapak Liman against Streptococcus mutans. Streptococcus mutans can make the extra-cell polysaccharide consists mainly of a glucose polymer that forms a plaque matrix and can stick on the surface of the tooth causing plaque buildup and bad breath. This research was purely experimental by made the Tapak Liman leaves into a viscous extract, then it was made as the antiseptic mouthwash and testing of its inhibiting power against Streptococcus mutans. The antiseptic mouthwash of leaf extract of Tapak Liman is made into five formulas consist of F0 (base), F1 (10%), F2 (20%), F3 (30%) and Fp as comparator (Betadin mouthwash). The antibacterial activity test of the antiseptic mouthwash of Tapak Liman with the diffusion method using the Nutrient Agar media (NA) and data was analyzed by One Way ANOVA. The result of research that the leaf extract of Tapak Liman in 3 concentrations variance can be formulated into the antiseptic mouthwash. The antiseptic mouthwash provides an average diameter – the average resistance of the barrier is F0 (8.40 mm), F1 (12.40 mm), F2 (12.80 mm) and F3 (13.60 mm) and Fp (14.80 mm). Conclusion of research that all of the formula leaf extract of Tapak Liman can be made as the antiseptic mouthwash, and all formulas that give inhibiting power test against the Streptococcus mutans

Keywords: Antiseptic mouthwash, extract, Tapak Liman leaves, Streptococcus mutans

INTRODUCTION

Indonesia's natural wealth, especially plants, has long been utilized by Indonesian people as a traditional medicinal ingredient. Some are still based on hereditary experiences and others have been developed through scientific research (Hariana, 2008).

One of the herbs that can be used as medicine is the Tapak Liman (*Elephantopus scaber L*). Tapak liman leaves efficacious as an antibacterial because it contains phenol, flavonoids and saponins compounds, also potentially as antibacterial because of its flavonoids content (Yuniarti, 2008). The highest content found on Tapak Liman leaves is a flavonoid, which is an antimicrobial that can inhibit the growth of gram-positive and gram negative bacteria e.g. *Streptococcus viridans* and *Streptococcus mutans* which cause dental problems and bad breath.

Halitosis is a problem in the mouth of unpleasant breath that comes out of the oral cavity. Halitosis is one of the problems of oral health that is much complained of society after the caries and periodontal disease (Wijayanti *et al*., 2010). Halitosis arises because of bacteria that live normally on the surface of the tongue and in the esophagus. These bacteria are normally there because they help the human digestive process by breaking down proteins. Bacteria species that are found on the oral surface can be sacramolithic, i.e. using *carbohydrates* as an energy source. Other species are either *asakarolytic or proteolytic*, i.e. using proteins, peptides or amino acids as their primary source. Most gram-positive bacteria are *sacramoolithic* and the gram-negative bacteria are *asakarolytic or proteolytic*. Gram-negative bacteria are the primary inhabitants of the supragingival plaque including plaque covering the tongue and other mucous surfaces (Oeding et al, 2012).

One way to overcome the smell of mouth is by using mouthwash that contains antibacterial substances. The antibacterial mouthwash is a formula of

solution, generally in concentrated form that must be diluted before use, intended to be used as a prevention or treatment of throat infections. According to other definitions, bottle is a solution that usually contains a breath freshener, astringent, demulsen or surfactant, or antibacterial to freshen and cleanse the respiratory tract with gargle (Akarina, 2011).. *Streptococcus mutans* is a bacterial cause of bad breath (Saswati & Indranil, 2011).

In this study carried out test of the antibacterial mouthwash activity of leaf extract of Tapak Liman (*Elephanthopus scaber L*) against the growth of *Streptococcus mutans bacteria*, using the diffusion method. In the Diffusion method to determine the activity of antimicrobial agents, using a disc paper that contains antimicrobial agents placed on blood media so that has been planted microorganisms that will diffuse in the blood media to the clear areas indicate that there is a growth barrier of microorganisms by antimicrobial agents on the surface of Agar nutrient (Pratiwi, 2008). Research objectives as follows: To find out the Tapak Liman leaf extract formula that can be made as a mouthwash. To know the power test of Tapak Liman leaf extract mouthwash against the bacteria *Streprococcus mutans*. The benefits of this research are as follows: Create the antibacterial mouthwash that comes from natural ingredients.

The product of Tapak Liman leaf extract mouthwash can be used by the community as a new mouthwash that efficacious as an antibacterial, preventing bad breath and cavitative tooth disorders. This antiseptic mouthwash product can be traded to increase public income.

PURPOSE OF RESEARCH

The purpose of this research to know the formulation of the leaf extract of Tapak Liman which can be made as a antiseptic mouthwash and know the inhibiting power test of the leaf extract of the Tapak Liman against Streptococcus mutans

METHODS

This type of research is purely experimental studies, by means of making the antiseptic mouthwash of leaf extract of Tapak Liman (*Elephantopus scaber L*). The antiseptic mouthwash is tested for its inhibiting power against *Streptococcus mutans*, with methods of diffusion and the use of Nutrient Agar-mediated (NA)

Tools and Materials

Tools

The tools used in this research are autoclaves (WEBECO), Petri Bowls (Pyrex), incubators (Memmet), Blender, wheelbarrow, round ose, pH indicator paper, LAF (Laminar Air Flow), Bunsen, micropipettes, Ovens (Memmet), tweezers, rotavators, scales Analytic (Mettler), scale pipettes, horn horns, spotes, teasers, refrigerators, tube racks, wheeled frames and laboratory glassware.

Materials

The materials used in this research are, Tapak Liman leaves (*Elephantopus scaber L*), aquades, ethanol 70%, medium NA (Nutrient Agar), Menthol, glycerin, Na-sakarin, label paper, filter paper, aluminium foil, and *Streptococcus mutans* bacteria.

Working procedures

Sample

Tapak Liman leaves (*Elephantopus scaber L*) were taken from Limau Manis area, Padang district, West Sumatra Province, Indonesia Country.

Sample Extraction

Wet samples were washed clean then dried by way of blowing air until dry without using direct sunlight then dragged into a smooth degree according to the subtle degree that is common for simplisia. (Yuniarti, 2008).

A total of 1500 grams of simplisia was then macerated in ethanol 70%. Maceration was carried out for 3 days while occasionally stirring. The maserate is then filtered to separate the ethanol fluid. Performed remaceration for 3 days then done filtering. The maceration was then incorporated into the rotary evaporator to obtain a condensed extract (Yuniarti, 2008).

The antibacterial mouthwash was made in several formulas that can be seen on;
The formulation of antibacterial moutwash of leaf extract of Tapak Liman

Ingredients	Formula of Antibacterial Mouthwash of leaf extract of Tapak Liman			
	F 0	F1	F2	F3
Extract (%)	0	10	20	30
Gliserin (%)	15	15	15	15
Propilenglikol (%)	0,15	0,15	0,15	0,15
Menthae piperitae(%)	0,30	0,30	0,30	0,30
Na. sakarin	0,25	0,25	0,25	0,25
Etanol (%)	0,05	0,05	0,05	0,05
Aquadest sampai (ml)	100	100	100	100

Preparation of antiseptic mouthwash

The antibacterial mouthwash solution is made by mixing all the ingredients and the ad aquadest 100 ml. Homogenized with magnetic stirrer speed of 100 rpm for 15 minutes. The prepared mouthwash solution is inserted into the tightly sealed container and stored in a cool place to be evaluated (Akarina, 2011).

Organoleptical Evaluation

Evaluation of antibacterial mouthwash is done by observing in terms of taste, shape, color, aroma and clarity. This examination is conducted at room temperature (15 – 30 º C) weekly for 6 weeks.

Preparation and inhibiting power test of Streptococcus mutans bacteria

Bacterial intake

Streptococcus mutans bacteria is obtained from the Microbiology Laboratory of the Faculty of Medicine of Andalas University Padang.

Bacterial rejuvenation

Streptococcus mutans bacteria derived from their mulberry

As much as 1 ose is then grown or inoculated by the way it is scratched in the medium Nutrient Agar (NA) tilt. The bacterial culture in each of them was incubated at a temperature of 37^{o}C for 18-24 hours. (Pratiwi, S., 2008)

Manufacture of Bacterial Suspension

Streptococcus mutans derived from their Mulberry origins are taken as much as 1 ose, then grown or inoculated by the way the Nutrient Agar (NA) tilts. The bacterial culture in each of them was incubated at a temperature of 37^{o}C for 18-24 hours. (Pratiwi, S.,2008)

Inhibiting Power Test of Antiseptic Mouthwash of Tapak Liman (*Elephantopus scaber L*)

Testing antibacterial activity using the diffusion method with the media Nutrient Agar (NA) as media through the observation of large diameter of the area, the media that has been sterilised, Nutrient Agar (NA) is poured into a petri dish ± 20 ml, after the media dipped Cotton Lidi sterile into the bacterial suspension, then wiped evenly on top of the media, subsequent sterile disc paper is transmitted with a 10 ml of mouthwash is then incubated at a temperature of 37^{o}C, for ± 24 hours. Observed the diameter of the inhibiting power is characterized by the presence of clear areas of signs not overgrown by bacteria. Testing was conducted against the preparation of F1, F2, F3 and as a negative control of the base of the antiseptic mouthwash, the same Fp as comparator (Betadin mouthwash).

Discussion

From the results of organoleptic evaluation which includes the examination of form, smell, color and flavor, for 4 weeks there is no change during storage. This indicates that the preparation of the mouthwash is stable during physical storage (Fig. 4.1).

Figure 4.1. Dosage Formula for the antibacterial mouthwash

Description:
F0 = Formula of antibacterial mouthwash with the concentration 0% of Tapak Liman leaf extract
F1 = Formula of antibacterial mouthwash with the concentration 10% of Tapak Liman leaf extract
F2 = Formula of antibacterial mouthwash with the concentration 20% of Tapak Liman leaf extract
F3 = Formula of antibacterial mouthwash with a concentration 30% of Tapak Liman leaf extract

Phytochemical testing gives the results that a positive the Tapak Liman leaves extract contains the compound of flavonoids, phenolic, and saponin, because it contains flavonoids then Tapak Liman leaves have antibacterial properties. Organoleptic test in terms of translucent texture and soft, clear brown color, mentha piperitae taste, menthae piperitae aroma, and a bit foaming.

The antibacterial activity test of the dosage of Tapak Liman leaves extract is done using the diffusion method by placing a sheet of sterile discs with a diameter of 5 mm and then tested with 10 ml of leaf extract of Tapak Liman on top of the Nutrient Agar media for which has been carried out bacterial suspension with sterile cotton lidi. The result (viewable in Table 4.2, Figure 4.2.) obtained the average resistance diameter at F3 = 13.60 mm, F2 = 12.80 mm, F1 = 12.40 mm Where these three formulas belong to the strong category. The average Diameter of the power is F0 = 8.40 mm which can inhibit the growth of bacteria and belongs to a strong group, because in F0 there are preservatives that can inhibit microbial growth. As a positive control the comparison is used "Betadine antiseptic mouthwash", where the average diameter of the power is 14.80 mm, which belongs to the strong group. It means that at a comparison dosage also has antibacterial activity against Streptococcus mutans.

Based on response table bacterial growth barriers according to Davis and Stout (1971) The power classification is divided into four categories: Strong = 10-20 mm, medium = 5-10 mm, weak = < 5 mm.

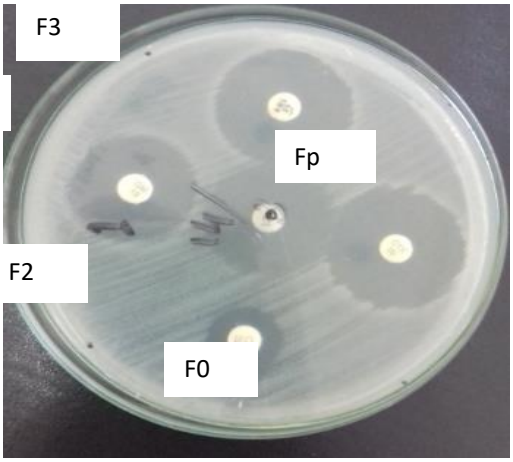

Figure 4.2. Activity antibacterial test of leaf extract of Tapak Liman as antibacterial mouthwash

Table 4.2. The formula of Tapak Liman leaves extract as antibacterial mouthwash in Diameter average of inhibiting Power test

Repetition	Diameter average of Inhibiting power test (mm)				
	F0	F1	F2	F3	Fp
1.	8,40	12,80	12,80	13,60	14,80
2.	8,80	12,00	12,40	13,30	14,80
3.	8,00	12,40	12,60	13,20	14,80
Average ± SD	8,40±0,5084	12,40 ±0,9792	12,80± 0,9641	13,60± 0,330	14,80± 0,8528

Tests of antibacterial activity were analyzed using ANOVA one-way statistics, obtained by the results that the concentration significantly affects the bacterial barrier with a value of < 0.05 P of 0.03.
The antibacterial mouthwash of Tapak Liman leaves extract have antibacterial activity against the growth of *Streptococcus mutans* so that it can be used to prevent and reduce bad breath.

Table. 4.3. Test One Way ANOVA Formula and bacterial barrier.

Daya Hambat Obat Kumur Ekstrak Daun Tapak Liman Terhadap Streptococcus mutans

	Sum of Squares	df	Mean Square	F	Sig
Between Groups	68,79	5	14.43	9.564	0.045
Within Groups	68	15	0.207		
Total	68.08	15			

The result of the formula F3 of inhibiting power test gives good from others. The results of this research can create a antibacterial mouthwash of leaf extract of Tapak Liman in the form of natural substances and cheap and easy to be taken from our environment.

CONCLUSION

The conclusion of research ; The leaf extract of Tapak Liman (*Elephantopus scaber L*) from 3 variations of concentration can be formulated in the form of antibactrial mouthwash. All of Tapak Liman in antibactrial mouthwash formula gives inhibiting power against *Streptococcus mutans* bacteria and formula F3 provides the greatest resistance to the growth of Streptococcus mutans, which is 14.60 mm almost identical to Fp as a comparator (Betadine mouthwash).

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